

UNIVERSITY OF CALIFORNIA, RIVERSIDE

BERKELEY • DAVIS • IRVINE • LOS ANGELES • MERCED • RIVERSIDE • SAN DIEGO • SAN FRANCISCO



NTA BARBARA • SANTA CRUZ

Graduate Program in Environmental Toxicology

University of California
Riverside, CA 92521
Telephone: (951) 827-4116

COLLEGE OF NATURAL AND AGRICULTURAL SCIENCES

July 31, 2008

To: Gerald W. Bowes, Ph.D.
Staff Toxicologist (Sup.)
Manager, Toxicology and Peer Review Section
Division of Water Quality
State Water Resources Control Board
1001 I Street
Sacramento, CA 95814

RECEIVED

AUG 05 2008

DIVISION OF WATER QUALITY

From: Dr. David A. Eastmond
Telephone: (951) 827-4497
FAX: (951) 827-3087
e-mail: david.eastmond@ucr.edu

A handwritten signature in cursive script that reads 'David A. Eastmond'.

RE: Review of the draft trichloropropane document

Dear Gerald:

Attached is a printed copy of my review of the draft document entitled, "Public Health Goal for 1,2,3-Trichloropropane in Drinking Water". Please let me know if you have any questions.

Thanks,

Dave

Review of the Draft Report Entitled "Public Health Goal for 1,2,3-Trichloropropane in Drinking Water".

by

Dávid A. Eastmond, Ph.D.
Environmental Toxicology Graduate Program
Department of Cell Biology & Neuroscience
University of California, Riverside

July 31, 2008

Overview:

Based on a review of the literature, OEHHA has concluded that 1,2,3-trichloropropane (TCP) should be considered a carcinogen for the development of a health-protective level in drinking water. This conclusion was based primarily upon the results of animal cancer bioassays, although information on TCP's genotoxicity, mechanism of action, metabolism, and pharmacokinetics was also considered. To calculate potency and derive a proposed public health goal (PHG), the relationship between dose and response in the low dose region was estimated from the NTP's 2-year animal cancer bioassay using a multistage Weibull time-to-tumor model to establish an LED₁₀ and then applying linear extrapolation to estimate low dose carcinogenic effects. For non-cancer effects, a health protective acceptable daily dose of 80 ppb was determined using a NOAEL with a series of uncertainty factors. The final proposed PHG of 0.0007 ppb, derived using the multistage Weibull time-to-tumor model combined with linear extrapolation, was calculated based upon an increase in forestomach tumors seen in female B6C3F1 mice administered TCP by oral gavage. The proposed PHGs for both non-cancer and cancer effects have been derived using commonly used methods and risk assessment techniques. My comments on the draft document including the requested questions are presented below.

Specific questions:

- 1) Accuracy of the information presented, including data on toxicity, metabolism, mode(s) of action and exposure, including the potential for carcinogenicity and reproductive toxicity.

2
The information presented appeared to be accurate in the majority of the areas that I checked. I did notice a number of relatively minor errors, which are listed in the technical comments section below. There are also a few potentially relevant studies that are not cited in the document. These are also listed in the technical comments section. For a number of the sections, there appears to be a strong reliance on secondary sources (ATSDR, WHO, etc.) and it was not clear if the primary sources had been checked. The reliance on secondary sources is a particular problem when using the HSDB which generally just copies and pastes things from other sources. In cases such as in Table 1 where the secondary sources provide somewhat differing values, it is not clear why one value was selected and not the other (see technical comments below). Unless there is a compelling reason to choose one over the other, I would recommend providing both values.

3
There seem to be inconsistencies in the Production and Uses section. Usage for TCP is listed at >1 million pounds per year and yet there is very little if any exposure data. If the usage occurs primarily or exclusively within a contained system, this should be stated. Additional and informative details on the production and use of TCP are provided in the WHO CICAD document (WHO, 2002). From the WHO document, it appears that the largest production of TCP occurs as a by-product during the manufacture of epichlorohydrin. I would recommend that this information be included in the PHG document. Additionally, it appears that one way by which TCP could get into the water supply would be through its presence as a by-product in pesticides and fumigants. While this is briefly mentioned, I would suggest that additional details be provided such as those described in WHO (2002) [e.g. TCP presence within Telone at 0.17% by weight; its presence within epichlorohydrin, etc.].

3
I find the metabolism data to be somewhat confusing. The abbreviations listed in Figure 1 should be defined. [Actually, I would recommend providing the names of the metabolites in the figure as has been done in the recent EPA IRIS TCP document.]. The metabolite names used in the text appear to differ from those used in the Figure. This is confusing and should be changed so that the metabolite and conjugate names in the text match those used in the figure. The metabolic details are hard to follow given the use of various inducers,

10-4
inhibitors, endpoints, etc. I would suggest that a brief summary of the critical points be presented at the end. From my reading, it appears that TCP requires bioactivation to exert many of its toxic effects. The data appear to indicate that cytochrome P450 monooxygenase-mediated activation can result in metabolites that primarily bind to proteins whereas bioactivation involving glutathione results in metabolites that preferentially bind to DNA. If this is a correct interpretation then I would suggest that this be included in the summary and that the Mechanism section on pages 20-21 be revised to be more informative.

C S
With regard to the mode of action, there is clear evidence that TCP is mutagenic and genotoxic in *in vitro* systems. In older *in vivo* studies, TCP was reported as negative for the induction of dominant lethal mutations in one study (Saito-Suzuki et al., 1982) but positive for altering hepatocyte ploidy levels (e.g. polyploidy) in two studies (Belyaeva et al., 1974 and 1977). [The citations can be found in EPA (2007) and should probably be included in this document.] TCP has also shown positive results in a number of studies of DNA adducts and strand breaks. Overall, these results indicate that TCP is likely to be mutagenic *in vivo* as well. The detection of uncommon and specific point mutations in *H-* or *K-ras* in the tumors of TCP-treated animals also provides persuasive evidence that TCP acts through a mutagenic mode of action. In my opinion, the high incidence and specificity of the mutations reported by Ito et al. (1996) should be emphasized rather than the observation that the detected transversions were not consistent with the authors' predicted miscoding properties of the adduct.

C C
I think that the mechanistic aspects of the document would be strengthened by adding a short section describing the results seen with other short-chain halogenated alkanes. Information on 1,2-dibromo-3-chloropropane (DBCP) would appear to be particularly relevant as the same DNA adduct is apparently formed by both TCP and DBCP [see WHO (2002), NTP (1993) and EPA (2007) for more information].

C 7
There is some information on immunotoxicity in the ATSDR monograph (ATSDR, 1992). In addition, in a PubMed search, I was able to identify one article [Albrecht WN (1987) J Toxicol Environ Health 21:405-21], which according to the abstract, may provide some

information on the neurotoxicity of TCP. If relevant, OEHHA may want to incorporate this information into the respective sections on page 14 of the document.

2) The appropriateness of the approach and studies used in developing the draft PHG, including selection of the data set and supporting information, particularly regarding interpretations of carcinogenicity data and mechanisms.

C8 A variety of approaches were applied to derive the proposed PHG for both cancer and non-cancer effects. These ranged from the conventional NOAEL/UF approach to multistage Weibull modeling combined with linear extrapolation into the low dose region. The approaches used are generally accepted and appear to be appropriate. Indeed, the approach used in this document to estimate the cancer effects is notably similar to that being proposed by the EPA IRIS program for TCP. [There are also differences but the overall approach is quite similar.] For the non-cancer effects, I was a bit surprised that the newer benchmark dose (BMD) approach was not used, but the use of the NOAEL is certainly acceptable.

C9 For assessing the non-cancer effects, OEHHA chose hematological changes occurring in the 17-week NTP study as the critical endpoint and study. While this seems to be an acceptable choice, it is not clear why they chose this study and endpoint rather than other changes such as the renal or hepatic changes that appeared in the 2-year bioassay. The rationale for the selection of this endpoint and study should be provided. The statement on page 21 that "no toxic effects that were not related to the occurrence of tumors were identified in the subsequent two-year cancer bioassay" seems questionable to me, given some of the changes reported (e.g a high incidence of hyperplasia in the kidney without an increase in kidney tumors, an increase in eosinophilic foci in the liver, etc.). It should be noted that in their recent evaluation the EPA discounted the hematological changes and based their reference dose on liver and kidney changes seen in the 2-year bioassay.

For generating the cancer risk estimates, the data from the NTP study was selected as increased incidences of tumors were seen in multiple tissues in both the male and female mice and rats. OEHHA modeled the tumorigenic responses in a number of tissues in both

sexes and species. For deriving the LED₁₀ and for extrapolation into the low dose region, the authors selected to model tumors that developed in the forestomach of female mice. The incidence of tumors was very high in these animals with 23 of 50 mice in the main study exhibiting squamous cell papillomas and 46 of 50 exhibiting squamous cell carcinomas at the lowest dose tested (6 mg/kg bw). As a result, very little information is available about the shape of the dose response curve below this dose in this sex and species. Because of this, the EPA (2007) chose not to use the mouse data for their modeling studies and instead used the rat studies where additional lower dose response information was available.

C10 I do have a recommendation for the presentation of the animal tumor data. The data presented in Table 4 and 5 presents only the tumor incidence for animals in the main NTP study. However, in some cases including the modeling used to estimate the cancer potency estimates, animals from the interim sacrifice group were combined with those of the main study for the analysis. The data from both the main and interim studies should be included in Tables 4 and 5. I might note that combining the interim sacrifice animals with those of the main study has also been done by the EPA (2007) and by the NTP (Irwin et al, 1995) when they presented or evaluated the TCP data. In addition, the doses used for the bioassay should be included in Tables 4 and 5. When feasible, I would suggest presenting the data in three categories: papillomas, carcinomas, and papillomas or carcinomas. Since many of the mice had more than one tumor, it is not clear to me how this was handled when modeling the data.

C11 OEHHHA also mentions that animals from the interim sacrifice group were censored (removed from the analysis if they did not display a tumor because they did not live until the end of the animal's natural lifetime. While this might be warranted for some of the tumors observed, it does not seem warranted for the mouse forestomach tumors given the high incidence of forestomach tumors in the interim sacrifice animals. Indeed this would seem to me to bias the results. This should be either changed or more effectively justified. The actual female male forestomach data used for the critical analysis should be presented.

3) Data evaluation and interpretation. Identification of key studies and the use of animal and human data in dose-response assessment. Do the data support the conclusions?

As indicated above, the 17-week and the 2-year NTP animal bioassays were selected as the key studies for deriving the proposed PHG. The 2-year NTP bioassay is clearly the best choice to evaluate the carcinogenicity of TCP. The choice of the 17-week study for the non-cancer effects seems to be an acceptable choice but should be justified as other studies such as the 2-year bioassay or the reproductive studies could have been chosen.

TCP is clearly a strong and potent animal carcinogen in animals and there is no adequate data on humans. As a result, it would seem biologically plausible and prudent to proceed as if it poses a carcinogenic risk to humans.

4) The appropriateness of the risk assessment methodology used. Are there other better methods that might improve the risk estimate?

The risk assessment methods that have been used are generally accepted and would appear to be appropriate (with the possible exception of the issues mentioned above), particularly as key mechanistic information is unavailable. However, while standard health-protective methods have been used to estimate the risks in the low dose region, the resulting proposed PHG is so low (>1 ppt) that one can't help but wonder if the approach being used isn't overly conservative. The observation that seemingly minor assumptions can change the potency estimates in the time-to-tumor model by more than 100-fold is a concern. Indeed, the fact that so many of the major effects seen in the 2-year cancer bioassay were cancer-related would suggest to me that the assumption that the carcinomas and papillomas were the cause of death (footnote 4) might be a more likely option for the time-to-tumor modeling.

Because the CSF is derived from the LED_{10} , R on page 26 is really an upper estimate of the individual cancer risk. This should be presented as such here and throughout the document.

5) Other major and critical information that might affect the estimates of impacts on public health.

C15
C16
Currently, details about the mechanisms underlying TCP carcinogenesis are unknown and, as indicated above, a mutagenic mode of action seems likely. However, there is DNA adduct data that suggests that the relationship between dose and response may be influenced by the route of administration, and more be more related to peak dose rather than cumulative dose. To me, this also implies that the dose response relationship may have a non-linear component. In their studies, La et al. (1996) demonstrated that DNA adduct levels and cell proliferation in selected TCP target tissues were significantly higher when a 6 mg/kg dose of TCP was administered orally by gavage as compared to when it was administered via drinking water. This suggests that risk estimates based on the NTP studies, which were conducted by oral gavage, may overestimate the cancer risk of TCP present at low concentrations in drinking water.

6) The appropriateness of identifying and quantifying the uncertainties in the PHG calculation. Considering the uncertainties, does the conclusion provide adequate public protection?

C17
While there isn't a specific section that describes the uncertainties, I believe that most of the uncertainties can be identified in reading the document. I don't believe that a separate section for uncertainties is necessary but could be added if desired. I believe that the proposed PHG provides ample protection for public health.

Detailed Comments

- a. There are a number of errors in Table 1. The units for the density should be g/cm^3 rather than g/cm^2 . Based on the references cited, the solubility in water value should be 1750 mg/L. The values for a number of properties differ between the references cited. The HSDB lists the vapor pressure as 3.69 and the Henry's law constant as 3.43. The log Koc is listed in the ATSDR monograph as 1.99 and the log Kow as 1.98. As indicated above unless there is a compelling reason to list one or the other, I would list both. [For

an example, see the recent EPA IRIS documentation on TCP.].

b. pp. 3-4. I would recommend that the assumptions that were used to derive the persistence values (e.g. half lives) reported in the air, soil and water be provided.

c. p.4. The importance of volatilization as a loss process from the soil should be mentioned.

✓ d. p.4. Soil section. The last sentence is misleading. TCP was a contaminant in the nematocides that were used rather than actually being the nematocide.

✓ e. For Tables 2 and 3, I would suggest that the meaning of "sources" be provided in the text or in a footnote.

✓ f. In Table 3, I would recommend that <1 be used rather than the dash (-).

✓ g. p.6. It would be helpful for the reader if the radioisotopes that were used in the studies were listed.

✓ h. p.7 and elsewhere. The term "cytochrome P450 monooxygenases" should be used rather than simply "cytochrome P450".

✓ i. p.12. I would add the word "close" before "concordance" when describing the DNA adducts. This is closer to the WHO (2002) and the EPA (2007) interpretation of the data which I prefer.

✓ j. p.14. There is mention of several tasks in the developmental and reproductive toxicity section. However, the descriptions are incomplete and don't make much sense. These should either be described in more detail (c.f. EPA 2007, p.41) or be deleted.

✓ k. pp. 16-17. The quality of the figures in the document is quite poor. Better copies of these figures should be included.

✓ l. p.19. According to the EPA (2007) document, kidney toxicity has been seen as an acute toxic effect of TCP [see Lag et al. (1991) Chem. Res. Toxicol. 4: 528-534].

✓ m. p.20. Add generally between was and positive in the 2nd line of the genotoxicity section.

n. p. 21. There is no difference in the incidence of clitoral tumors in the rats administered the high and medium doses, particularly when the incidence of benign and malignant tumors are combined.

✓ o. p.21 and elsewhere. The NTP subchronic study was conducted for 17 weeks rather than 90 days. This should be corrected throughout the document.

✓ p. p. 23. The potency estimate used for the low dose extrapolation should be more clearly indicated in Table 6.

- ✓ q. p. 23. Some of the q1* values in Table 6 seem unusually large and should be checked for accuracy.
- ✓ r. p.23. "Interim" sacrifice group should be used rather than "interval" sacrifice group.
- ✓ s. p.24. "for" should be deleted from the second line from the bottom.
- t. p.25. /LOAEL should be deleted from the ADD formula as this is describing what was done with this specific chemical.
- u. p.28. "AND GUIDELINES" should be added to the OTHER REGULATORY STANDARDS heading.
- v. p.28. (U.S.EPA, 2007; file last updated 08/01/1990) should be U.S.EPA, 2007b; file section last updated 08/01/1990).